

The exotic *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae) and other coccinellids in selected vineyards of the Western Cape Province, South Africa

K.A. Achiano^{1*}, J.H. Giliomee², E. Allsopp¹ & L. Williams¹

¹ARC-Infruitec-Nietvoorbij, Private Bag X5026, Stellenbosch, 7599 South Africa

²Centre for Invasion Biology, Department of Botany & Zoology, Stellenbosch University, Private Bag X1 Matieland, 7602 South Africa

Harmonia axyridis (Pallas) (Coleoptera: Coccinellidae) is an exotic ladybird, first recorded in South Africa in 2004, where it was introduced factitiously. In this study the seasonal occurrence of *H. axyridis* and other coccinellids were monitored in four vineyards and the adjacent windbreaks over three growing seasons in the Western Cape Province, South Africa. *Harmonia axyridis* made up the highest mean number of coccinellids collected per site over the collecting period, followed by *Hippodamia variegata*, *Cheilomenes lunata*, *Exochomus flavipes* and *Micraspis* sp. No indigenous parasitoids were observed attacking *H. axyridis*. The numbers of *H. axyridis* on the vines were negatively correlated with temperature but positively with rainfall. In view of the migration of *H. axyridis* from the vines in December, they appear not to pose any risk of contaminating grapes during harvest and tainting wine as most wine grapes are harvested from January onwards.

Key words: *Harmonia axyridis*, coccinellids, ladybirds, grapevines, abiotic factors.

INTRODUCTION

The intensification of human transport and commerce around the world has led to widespread movement of insect species outside of their native range (Mack *et al.* 2000; Hulme 2009; Tatem 2009; Giliomee 2011). Vitousek *et al.* (1996, 1997), Pimentel *et al.* (2000) and Simberloff (2004) considered such human-assisted invasions to pose a major threat to biodiversity worldwide, causing, among others, the displacement (Simberloff & Stiling 1996) and extinction (Williamson 1996; Evans 2004) of indigenous species.

Harmonia axyridis (Pallas) (Coleoptera: Coccinellidae) is a ladybird indigenous to Asia (Komai 1956) and its distribution extends across southern Siberia from the Altai Mountains to the Pacific coast, including Korea, Japan, southern China and the Himalayas (Dobzhansky 1933; Osawa 2011). It has in recent years emerged as a significant invader on several continents (Koch *et al.* 2006). It was introduced into North America (Oregon, U.S.A.) between 1970 and 1980 (Nalepa *et al.* 1996; Chapin & Brou 1991; LaMana & Miller 1996), South America (Argentina) in 2001 (Saini 2004), Europe (Great Britain) in 2004 (Majerus *et al.* 2006) and Africa (South Africa) in 2004 (Stals & Prinsloo 2007).

Harmonia axyridis thrives in temperate and sub-

tropical climates (Poutsma *et al.* 2008). It is also adapted to a wide range of temperatures from below 0 °C (LaMana & Miller 1998; Watanabe 2002) to 34 °C (LaMana & Miller 1998). At temperatures near 27 °C, development from egg to adult requires about 18 days. The developmental rate increases with increase in temperature (LaMana & Miller 1998).

The ladybird preys voraciously on a diversity of pestiferous aphids (Hukusima & Kamei 1970; Lou 1987; Hu *et al.* 1989; Lucas *et al.* 1997). It is polyphagous (Adriaens 2003) and will also attack soft-bodied arthropods when aphids are in short supply (Brown *et al.* 2011) and will also feed on other beneficial insect species (Yasuda & Ohnuma 1999), such as eggs and larvae of the ladybird *Exochomus flavipes* (Coleoptera: Coccinellidae), which preys on vine mealybugs (Achiano, pers. obs.). Stuart *et al.* (2002) also found that *H. axyridis* feeds rapaciously on the snout beetle *Diaprepes abbreviatus* (Coleoptera: Curculionidae) in Florida, U.S.A.

Harmonia axyridis can have a negative impact on wine making when it aggregates in clusters of grapes and is processed with the grapes, resulting in tainted, unmarketable wine or juice. The taint could result in undesirable peanut, bell pepper and asparagus aromas and flavours in wine

*Author for correspondence. E-mail: achianok@arc.agric.za

(Pickering *et al.* 2005). The deleterious effect on final wine quality increases with increasing numbers of *H. axyridis* processed with the harvested grapes (Pickering *et al.* 2004). Kovach (2004) and Pickering *et al.* (2007) determined the threshold density for wine contamination to be about 0.9 and 1.5 beetles/kg of grapes, respectively, above which interventions in the field or in the winery should be considered. The unacceptable taste associated with *H. axyridis* could cause severe losses to the wine industry in the Western Cape Province, as had happened in Canada and the U.S.A. (Pickering *et al.* 2004).

The aims of this study were to determine: (i) the seasonal abundance of *H. axyridis* in four selected vineyards in the Western Cape winelands, (ii) whether indigenous parasitoids that attack local predatory ladybirds also attack *H. axyridis*, (iii) the effect of abiotic factors such as temperature and rainfall on the numbers of *H. axyridis*, and (iv) whether it poses a risk to the local wine industry.

MATERIAL AND METHODS

Four vineyards (about 3 ha each), with pine tree windbreaks on at least one side were selected as trial sites, after preliminary investigation indicated the presence of *H. axyridis* on them. These were at Nietvoorbij (33°91'S 18°85'E; altitude 149 m), Glen Helderberg (34°03'S 18°42'E; altitude 100 m), Backsberg (33°83'S 18°91'E; altitude 242 m), and La Motte (33°83'S 18°61'E; altitude 248 m). On Nietvoorbij and Glen Helderberg chlorpyrifos was applied prior to bud burst to control overwintering populations of grapevine mealybug, *Planococcus ficus* (Signoret), but no mealybug control was applied on Backsberg and La Motte. Triticale was planted as a cover crop in April between the vine rows at all four sites and left to dry and die off at the end of August to September throughout the research period.

Each trial vineyard was divided into three blocks of approximately 1 ha each. In each hectare block, 20 plots of five vines each were randomly selected and the number of *H. axyridis* and other coccinellids on half of each vine recorded every two weeks. Visual inspection was also used to monitor the presence of *H. axyridis* and other coccinellids on the adjacent pine trees.

The feasibility of using sticky traps to monitor the occurrence of *H. axyridis* and other coccinellids in vineyards and adjacent pine trees was investi-

gated. One yellow sticky trap (Agribiol® 200 mm × 100 mm) was hung at a height of 1.0 m in each of the three blocks per vineyard. One yellow sticky trap was hung from a branch 1.0 m above ground in the adjacent pine tree windbreak corresponding to each hectare block in which a sticky trap was hung. Yellow delta traps™ (110 mm × 200 mm × 280 mm) containing white sticky pads were also hung at a height of 1.0 m and at a density of one trap/ha block. The yellow delta traps and yellow sticky traps were spaced 30 m apart in the same vine row in the centre of each hectare block to avoid potential edge effects. Traps were inspected fortnightly from September 2009 to March 2012. At every inspection the yellow sticky traps and the removable sticky inserts of the delta traps were replaced. The used traps were taken to the laboratory and inspected under a stereo-microscope to identify and count all the coccinellids.

Temperature and rainfall data were obtained from ARC-Institute for Soil, Climate and Water at Stellenbosch.

Statistical analyses

Pearson correlation of independent variables (temperature and rainfall) and dependent variable *H. axyridis* were calculated. Visual inspection data for the two sampling dates per month were combined to obtain the total number of *H. axyridis* counted per month in each of the four trial vineyards and the adjacent pine trees. The monthly total counts were averaged over three growing seasons (September 2009 to March 2012) and plotted against mean monthly temperature (°C) and mean monthly rainfall (mm), also averaged over three growing seasons. ANOVA was used to compare the mean number of each coccinellid species collected on four farms over three seasons. Factorial analysis was used to determine the interactions of year (Y), farm (F), and species (Sp.). All statistical analyses were performed on Statistica (2012), and treatment effects were considered significant at $P < 0.05$.

RESULTS AND DISCUSSION

The number of *H. axyridis* counted in vineyards peaked between October and December, and then declined, with one or two minor peaks between May and August, depending on the locality (Figs 1, 2). This pattern suggests multi-voltinism. *Harmonia axyridis* has been shown to be multivoltine, with

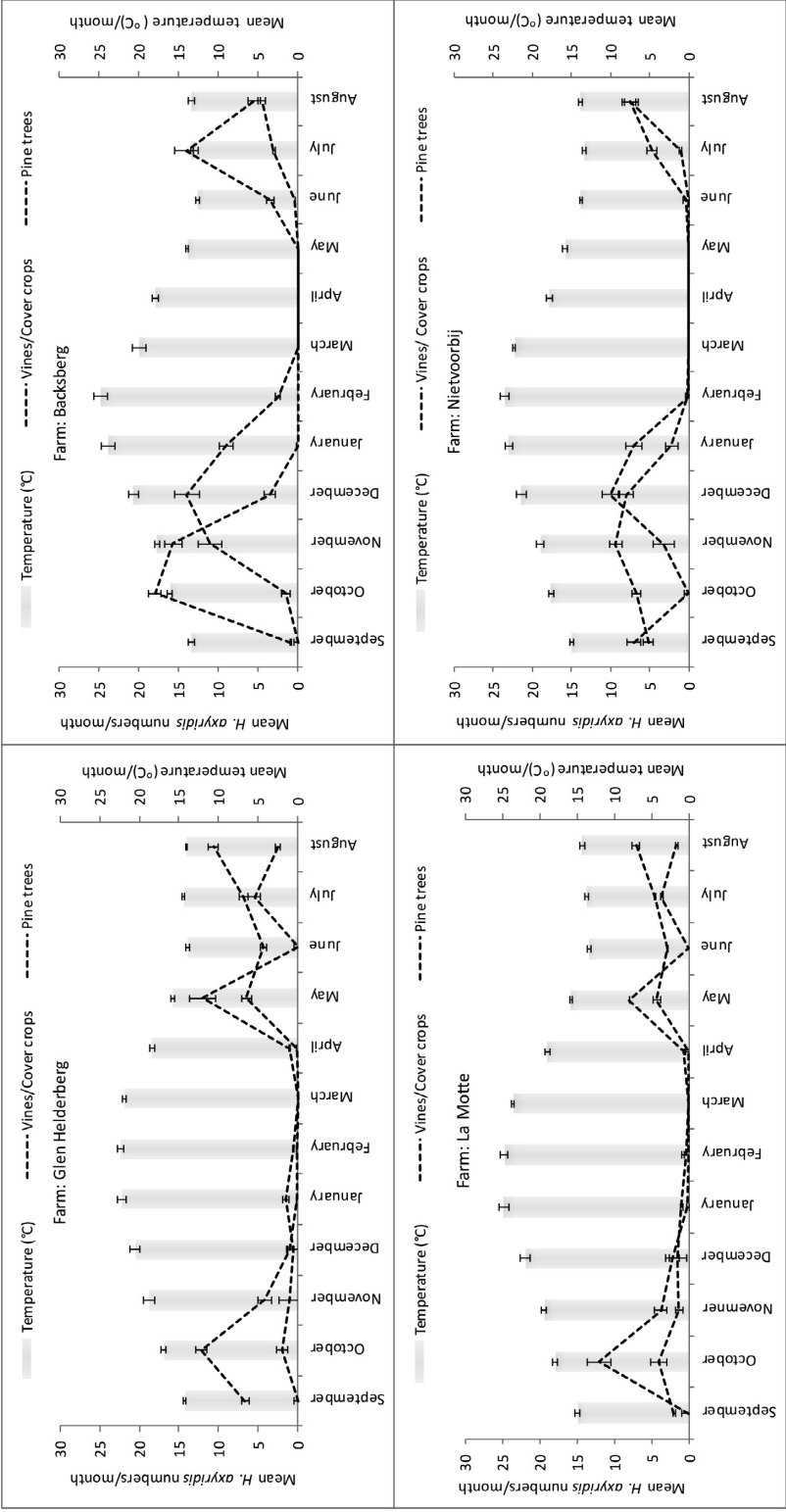


Fig. 1. Mean number of *Harmonia axyridis* counted per month in four Western Cape vineyards and the adjacent pine trees plotted against mean monthly temperature (°C) over three seasons (September 2009 to March 2012).

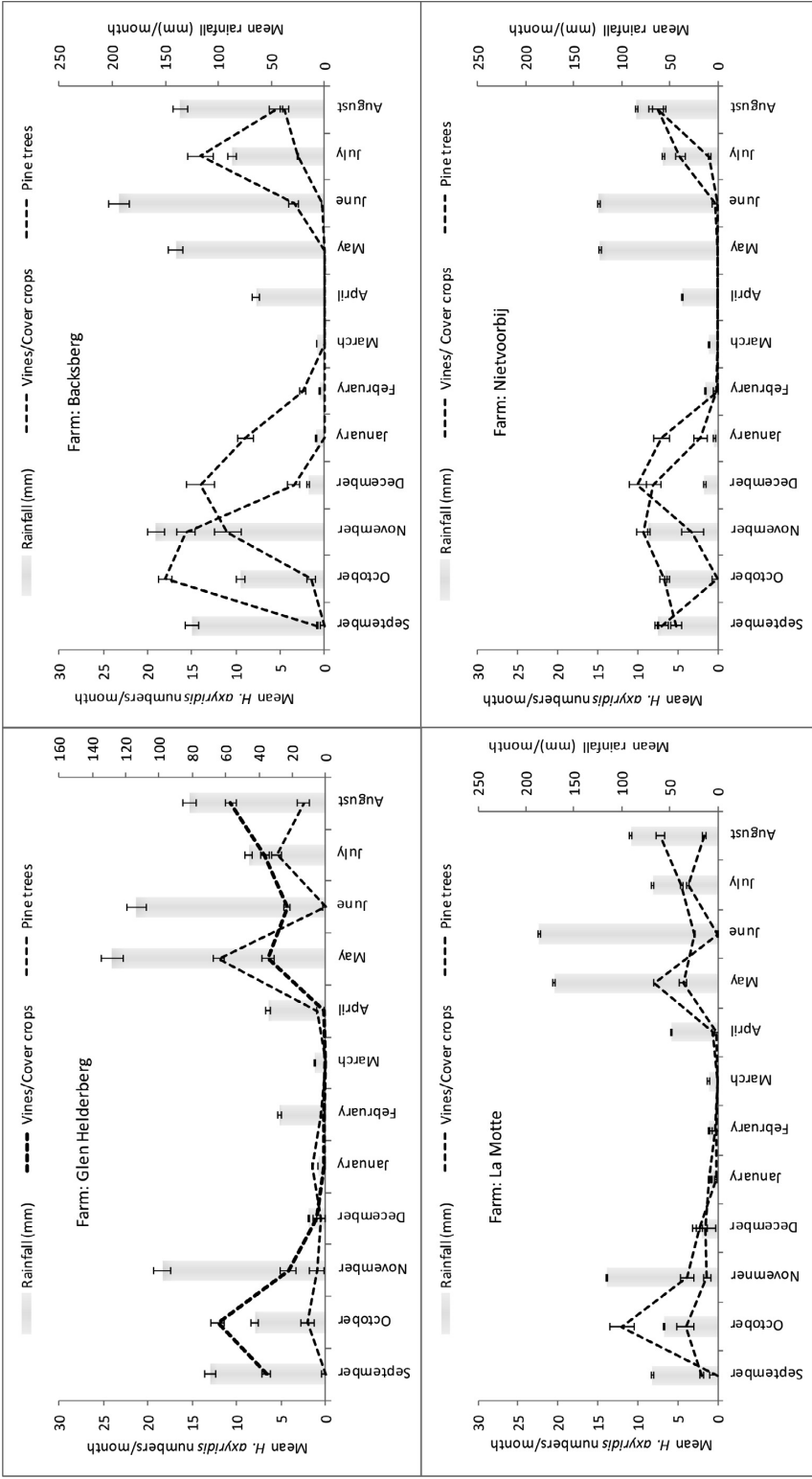


Fig. 2. Mean number of *Harmonia axyridis* counted per month in four Western Cape vineyards and the adjacent pine trees plotted against mean monthly rainfall over three seasons (September 2009 to March 2012).

four overlapping generations per year in Greece (Katsoyannos *et al.* 1997) and two in Japan and Great Britain (Osawa 2000; Brown *et al.* 2008). Multivoltinism in *H. axyridis* could be considered an advantage in terms of its population growth over the indigenous *E. flavipes* which is univoltine (Pretorius *et al.* 2010). The situation here is similar to that in Great Britain, where the dominant *H. axyridis* is multivoltine and the native coccinellids *C. septempunctata* and *Adalia bipunctata* (L.) are univoltine in most years (Majerus 1994; Brown *et al.* 2011).

The numbers of *H. axyridis* collected on the adjacent pine trees also showed an October peak on La Motte, but at Backsberg and Nietvoorbij numbers increased to a peak in December, while those on the vines declined (Figs 1, 2). The decrease of *H. axyridis* numbers on the vines in December and the increase on the pine trees coincided with an increase in temperature and a decrease in rainfall, respectively (Figs 1, 2). The Pearson correlation of beetle numbers on the vines with temperature and rainfall was -0.629 and 0.720 , respectively (at $P < 0.05$) (Table 1). Brown *et al.* (2011) also found numbers of *H. axyridis* to be negatively and positively correlated with temperature and rainfall, respectively. This was the period, from June to September, in which aphids were seen on the cover crops. Since the temperature and rainfall regime was the same in both the vineyards and surrounding

areas, the two abiotic factors could not be the driving factor for the emigration and immigration of *H. axyridis*. More likely it is the lack of food resources, such as aphids, that might have triggered the exodus of *H. axyridis* from the vineyards. *Harmonia axyridis* started moving out of the vines in mid-October and November when aphid numbers were decreasing as the cover crops were dying off. They returned in May when aphids began to colonise the next season's cover crops (Figs 1, 2). The application of chlorpyrifos to dormant vines did not seem to have an adverse effect on *H. axyridis*, since the reduction in numbers of *H. axyridis* was similar on the farms where no chlorpyrifos was applied (Figs 1, 2, 3). The quantification of aphid numbers could be useful in explaining the emigration and immigration of *H. axyridis* from the vineyards, and future work in this regard is recommended. The virtual absence of *H. axyridis* in vineyards during the harvesting season from January until April indicates that it does not pose any real risk of contaminating grapes during harvest and tainting wine.

The numbers of *H. axyridis* and other coccinellids caught with the yellow and Delta sticky traps were negligible (Table 2), indicating that these are not suitable for monitoring *H. axyridis* in vineyards.

The mean numbers of each coccinellid species counted over three growing seasons on grapevines and adjacent pine trees on four wine grape

Table 1. Pearson correlation of independent variables temperature and rainfall and dependent variable *Harmonia axyridis* numbers from all trial sites combined over three growing seasons.

Independent variables	Dependent variables	Pearson correlation	Significant difference ($P < 0.05$)
Temperature ($^{\circ}\text{C}$)	<i>H. axyridis</i>	-0.629	0.004
Rainfall (mm)	<i>H. axyridis</i>	0.720	0.002

Table 2. Mean number of each coccinellid species counted (caught with yellow and delta sticky traps) in four wine grape vineyards in the Western Cape Province over three growing seasons (September 2009 to March 2012).

Farm	Species				
	<i>H. axyridis</i>	<i>H. variegata</i>	<i>C. lunata</i>	<i>E. flavipes</i>	<i>Micrapis</i> sp.
Backsberg	0.04 (0.03) a	0.00 (0.00) a	0.06 (0.04) a	0.00 (0.00) a	0.00 (0.00) a
Helderberg	0.00 (0.00) b	0.02 (0.02) a	0.04 (0.03) bc	0.00 (0.00) b	0.00 (0.00) b
La Motte	0.04 (0.02) ab	0.07 (0.01) a	0.03 (0.03) a	0.00 (0.00) a	0.00 (0.00) c
Nietvoorbij	0.05 (0.03) a	0.00 (0.00) a	0.00 (0.00) a	0.00 (0.00) a	0.00 (0.00) a

Means with the same letters in a row are not significantly different ($P < 0.05$).

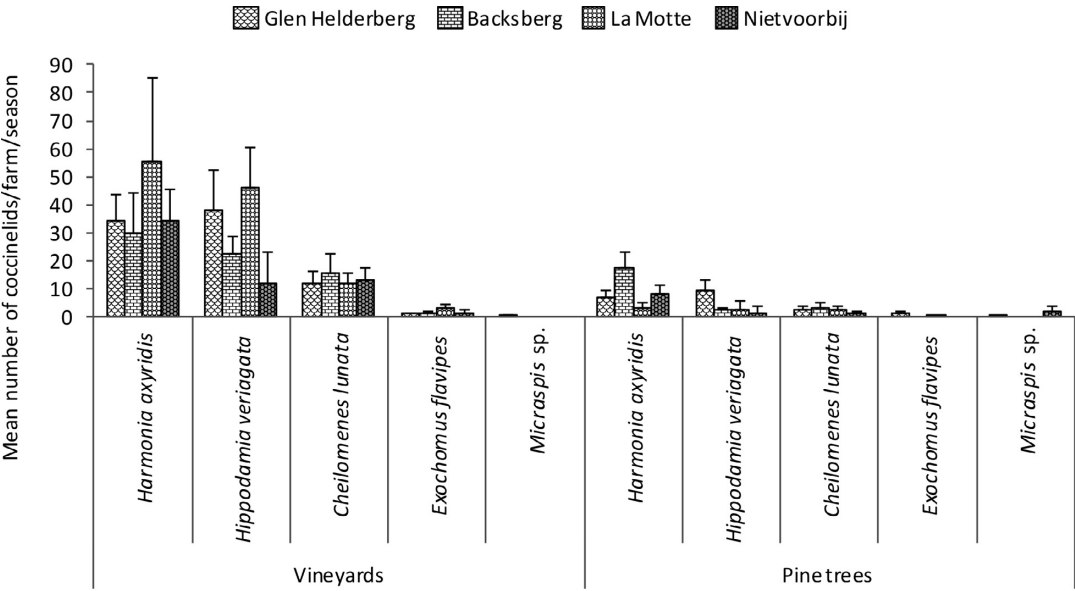


Fig. 3. Mean number of each coccinellid species counted over three growing seasons (2009 to 2012) on grapevines and adjacent pine trees on four wine grape farms in the Western Cape Province.

farms are presented in Fig. 3. The exotic *H. axyridis* was most abundant, with a mean of 55.10 counted on vines at La Motte (Fig. 3). In the adjacent pine trees numbers of *H. axyridis* were also found to be the highest, except at Glen Helderberg, where another exotic, *Hippodamia variegata* (Goeze), was most abundant. The means of all three seasons combined showed the relative dominance of *H. axyridis* (Table 3). With a mean of 40.67 it was significantly more abundant than *H. variegata* (32.15), *Cheilomenes lunata* (Fabr.) (12.84), *Exochomus flavipes* (Thunb.) (1.69) and *Micraspis* sp. (0.44) at $P < 0.05$. The dominance of *H. axyridis* over the indigenous coccinellids occurred over all three years of the survey (Table 3). Apart from a significant three-way interaction between Year, Farm and Species, the two-way interaction was

significant, implying that Farm and Year have a role in determining observed numbers for each species (Table 4). The interactions between Year and Species was highly significant, indicating the dominance of *H. axyridis* over the indigenous coccinellids. The dominant position reached in the short period since it was first observed in 2002 in South Africa, is indicative of the invasive nature of the species. This observation appears to be in line with the work of Brown *et al.* (2011) in England where *H. axyridis* numbers increased dramatically, whilst those of native aphidophagous species declined over a period of three years. Michaud (2002) also found that *H. axyridis* numbers increased, while those of the formerly dominant indigenous *Cycloneda sanguinea* (L.) (Coleoptera: Coccinellidae) declined over a period of five years after introduc-

Table 3. Mean number of each coccinellid species counted (by visual inspection) in four wine grape vineyards in the Western Cape Province over three growing seasons (September 2009 to March 2012).

Year	Species				
	<i>H. axyridis</i>	<i>H. variegata</i>	<i>C. lunata</i>	<i>E. flavipes</i>	<i>Micraspis</i> sp.
2009	30.38 (7.27) a	19.25 (3.74) b	15.94 (3.73) b	0.69 (0.28) c	1.19 (1.06) c
2010	52.19 (6.11) a	46.50 (7.80) a	14.13 (2.03) b	2.78 (0.98) c	0.06 (0.06) d
2011	39.44 (6.75) a	30.69 (3.37) b	8.44 (1.48) c	1.63 (0.65) d	0.06 (0.06) d
Mean of 3 years	40.67 (5.30) a	32.15 (3.18) b	12.84 (1.51) c	1.69 (0.41) d	0.44 (0.16) d

Means with the same letters in a row are not significantly different ($P < 0.05$).

Table 4. Analysis of variance for year, farm and number of species interactions.

Factor	d.f.	MS	F-value	P-level
Year (Y)	2	797.04	4.30	0.021
Farm (F)	3	692.04	3.73	0.020
Species (Sp.)	6	943.48	330.84	<0.0001
Y * F	6	726.10	3.92	<0.0001
Y * Sp.	12	601.08	21.08	<0.0001
F * Sp.	18	372.44	13.06	<0.0001
Y * F * Sp.	35	721.48	25.30	<0.0001
Error	195	5560.72	—	—

tion of *H. axyridis* into a Florida citrus ecosystem. *Harmonia axyridis* also appears to be relatively dominant over *H. variegata*, the other exotic coccinellid in South African vineyards (Fig. 1, Table 1). Colunga-Garcia & Gage (1998) found a similar phenomenon in the Michigan landscape where *H. axyridis* took the place of the exotic *Coccinella septempunctata* and became the dominant coccinellid in the ecosystem. However, in the absence of data on the abundance of the other coccinellids in vineyards where *H. axyridis* does not occur, the apparent displacement of other coccinellids by this invasive species in vineyards in the Western Cape must be regarded with some caution.

The ability of *H. axyridis* to dominate other species, even in their native range, has been attributed to various characteristics. Intra-guild predation could be one mechanism utilised by *H. axyridis* to displace indigenous species such as *E. flavipes* (Hautier *et al.* 2009). Evans (1991), Cottrell & Yeargan (1999), Yasuda & Ohnuma (1999) and Michaud (2003) also suggested that intra-guild predation might be facilitating the dominant invasion by *H. axyridis*. Studies done by Sakuratani *et al.* (2000), Kajita *et al.* (2000), Yasuda *et al.* (2001) and Koch *et al.* (2003) indicate that *H. axyridis* can effectively utilise other members of the aphidophagous guild as a food source. All the coccinellid species, indigenous and exotic, encountered in this study feed on aphids, thus at low densities of prey, *H. axyridis* may engage in intra-guild predation. It was shown that the intensity of predation by *H. axyridis* on other guild members is inversely related to aphid density (Hironori & Katsuhiro 1997; Burgio *et al.* 2002). Joseph *et al.* (1999) and Michaud (2003) found that *H. axyridis* has the innate ability to display kin recognition and is thus less likely to cannibalise a sibling than a non-sibling.

Harmonia axyridis females and other coccinellids refrain from laying eggs in the presence of an oviposition deterring semiochemical produced by *H. axyridis* larvae, thus reducing cannibalism and intraguild predation (Magro *et al.* 2007). This in part ensures its stranglehold as the top predator in the coccinellid guild. Takahashi (1989) and Lucas *et al.* (1997) suggested that the position of *H. axyridis* as a top intra-guild predator might be attributable to its aggressive nature and the shape of its mandibles, whilst Yasuda & Ohnuma (1999) attributed it to its ability to thrive on a varied diet, including other species of ladybirds.

Other factors such as high reproductive rate, good searching ability and two or possibly three generations per year could also contribute to the dominance of the exotic *H. axyridis*. Abdel-Salam & Abdel-Baky (2001) mentioned that *H. axyridis* has a higher capacity to multiply its population than other coccinellids, whilst Osawa (2000) and Yasuda *et al.* (2001) attributed the greater success of *H. axyridis* over other guild members to its higher attack rates and Yasuda *et al.* (2001) to greater escape ability. The relatively large size of *H. axyridis* (length 4.9–8.2 mm, width 4.0–6.6 mm) (Kuznetsov 1997) compared to *H. variegata* (length 4.4–5.0 mm, width 3.0–3.3 mm) (Gorden 1987) and other guild members could also account for the former's ability to displace other guild members. Majerus (1994), Lucas *et al.* (1997) and Michaud (2002) reported that in predatory interactions between coccinellids it is generally the larger that eats the smaller. *Harmonia axyridis* protects itself with a foul warning smell by secreting 2-alkyl-3-methoxypyrazines (Cudjoe *et al.* 2005; Cai *et al.* 2007) which makes it unpalatable or acts as an antifeedant to other predators of the coccinellid guild. In Japan, *H. axyridis* is unpalatable to *Coccinella septempunctata* (L.) (Yasuda *et al.* 2000).

Cheilomenes lunata, an indigenous coccinellid, is attacked by *Dinocampus coccinellae* (Schrank) (Hymenoptera: Braconidae) in South Africa (Ceryngier *et al.* 2012; Achiano, pers. obs.). However, no indigenous parasitoids were found to attack *H. axyridis* in this study and this will enhance its competitive edge over the indigenous coccinellids.

CONCLUSION

In view of the observed decline of *H. axyridis* numbers in vineyards from December onwards

when the cover crops and aphids have disappeared, it appears not to pose any risk of contaminating grapes during harvest and tainting wine, as most wine grapes are harvested from January onwards. *Harmonia axyridis* was consistently the most abundant coccinellid on grapevines in all four vineyards sampled over three growing seasons. No indigenous parasitoids were found to attack *H. axyridis*. It is possible that *H. axyridis* is displacing other exotic and indigenous coccinellids in

vineyards. However, it might also have filled a niche not occupied by the other coccinellids. This needs further investigation.

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REFERENCES

- ABDEL-SALAM, A.H., ABDEL-BAKY, N.F. 2001. Life table and biological studies of *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae) reared on the grain moth eggs of *Sitotroga cerealella* Olivier (Lepidoptera: Gelechiidae). *Journal of Applied Entomology* **125**: 455–462.
- ADRIAENS, T., BRANQUART, E. & MAES, D. 2003. The multi-coloured Asian ladybird *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae), a threat for native aphid predators in Belgium? *Belgian Journal of Zoology* **133**: 195–196.
- BROWN, P.M.J., ROY, H.E., ROTHERY, P., ROY, D.B., WARE, R.L. & MAJERUS, M.E.N. 2008. *Harmonia axyridis* in Great Britain: analysis of the spread and distribution of a non-native coccinellid. *BioControl* **53**: 55–67.
- BROWN, P.M.J., FROST, R., DOBERSKI, J., SPARKS, T., HARRINGTON, R. & ROY, H.E. 2011. Decline in native ladybirds in response to the arrival of *Harmonia axyridis* (Coleoptera: Coccinellidae): early evidence from England. *Ecological Entomology* **36**: 231–240.
- BURGIO, G., SANTI, F. & MAINI, S. 2002. On intra-guild predation and cannibalism in *Harmonia axyridis* (Pallas) and *Adalia bipunctata* L. (Coleoptera: Coccinellidae). *Biological Control* **24**: 110–116.
- CAI, L., KOZIEL, J.A. & O'NEAL, M.E. 2007. Determination of characteristic odorants from *Harmonia axyridis* beetles using in vivo solid-phase micro-extraction and multidimensional gas chromatography-mass spectrometry-olfactometry. *Journal of Chromatography* **1147**: 66–78.
- CERYNGIER, P., ROY, H.E. & POLAND, R.L. 2012. Natural enemies of ladybird beetles. In: Hodek, I., van Emden, H.F. (Eds) *Ecology and Behaviour of the Ladybird Beetles (Coccinellidae)*. Wiley-Blackwell, Chichester, U.K. 375–443.
- CHAPIN, J.B. & BROU, V.A. 1991. *Harmonia axyridis* (Pallas), the third species of the genus to be found in the United States (Coleoptera: Coccinellidae). *Proceedings of the Entomological Society of Washington* **93**: 630–635.
- COLUNGA-GARCIA, M. & GAGE, S.H. 1998. Arrival, establishment, and habitat use of the multi-coloured Asian lady beetle (Coleoptera: Coccinellidae) in a Michigan landscape. *Environmental Entomology* **27**: 1574–1580.
- COTTRELL, T.E. & YEARGAN, K.V. 1999. Intraguild predation between an introduced lady beetle, *Harmonia axyridis* (Coleoptera: Coccinellidae), and a native lady beetle, *Coleomegilla maculata* (Coleoptera: Coccinellidae). *Journal of the Kansas Entomological Society* **71**: 159–163.
- CUDJOE, E., WIEDERKEHR, T.B. & BRINDLE, I.D. 2005. Headspace gas chromatography-mass spectrometry: a fast approach to the identification and determination of 2-alkyl-3-methoxypyrazine pheromones in ladybugs. *Analyst* **130**: 152–155.
- DOBZHANSKY, T. 1933. Geographical variation in ladybeetles. *American Naturalist* **67**: 97–126.
- EVANS, E.W. 1991. Intra versus interspecific interactions of ladybeetles (Coleoptera: Coccinellidae) attacking aphids. *Oecologia* **87**: 401–408.
- EVANS, E.W. 2004. Habitat displacement of North American ladybirds by an introduced species. *Ecology* **85**: 637–647.
- GILIOME, J.H. 2011. Recent establishment of many alien insects in South Africa – a cause for concern. *African Entomology* **19**: 151–155.
- GORDEN, R.D. 1987. The first North American records of *Hippodamia variegata* (Goeze) (Coleoptera: Coccinellidae). *Journal of the New York Entomological Society* **95**: 307–309.
- HAUTIER, L., GREGOIRE, J.-C., CALLIER, P., SAN MARTIN, G., JANSEN, J.-P. & DE BISEAU, J.-C. 2009. Measuring the impact of *Harmonia axyridis* intraguild predation on native coccinellids in the field. Science Facing Aliens. *Proceedings of a Scientific Meeting on Invasive Alien Species* held in Brussels, 11 May, 2009. 31–34.
- HIRONORI, Y. & KATSUHIRO, S. 1997. Cannibalism and interspecific predation in two predatory ladybirds in relation to prey abundance in the field. *Entomophaga* **42**: 153–163.
- HU, Y.S., WANG, Z.M., NING, C.L., PI, Z.Q. & GAO, G.Q. 1989. The functional response of *Harmonia axyridis* to their prey of *Cinara* sp. *Natural Enemies of Insects* **11**: 164–168.
- HUKUSIMA, S. & KAMEI, M. 1970. Effects of various species of aphids as food on development, fecundity and longevity of *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae). *Research Bulletin of the Faculty of Agriculture, Gifu University* **29**: 53–66.
- HULME, P.E. 2009. Trade, transport and trouble: managing invasive species pathways in an era of globalization. *Journal of Applied Ecology* **46**: 10–18.

- JOSEPH, S.B., SNYDER, W.E. & MOORE, A.J. 1999. Cannibalizing *Harmonia axyridis* (Coleoptera: Coccinellidae) larvae use endogenous cues to avoid eating relatives. *Journal of Evolutionary Biology* **12**: 792–797.
- KAJITA, Y., TAKANO, F., YASUDA, H. & AGARWALA, B.K. 2000. Effects of indigenous ladybird species (Coleoptera: Coccinellidae) on the survival of a species in relation to prey abundance. *Applied Entomology and Zoology* **35**: 473–479.
- KATSOYANNOS, E., KONTODIMAS, D.C., STATHAS, G.J. & TSARTSALISM, C.T. 1997. Establishment of *Harmonia axyridis* on citrus and some data on its phenology in Greece. *Phytoparasitica* **25**: 183–191.
- KOCH, R.L., HUTCHISON, W.D., VENETTE, R.C. & HEIMPEL, G.E. 2003. Susceptibility of immature monarch butterfly, *Danaus plexippus* (Lepidoptera: Nymphalidae: Danainae), to predation by *Harmonia axyridis* (Coleoptera: Coccinellidae) *Biological Control* **28**: 265–270.
- KOCH, R.L., VENETTE, R.C. & HUTCHISON, W.D. 2006. Invasions by *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae) in the western hemisphere: implications for South America. *Neotropical Entomology* **35**: 3–4.
- KOMAI, T. 1956. Genetics of ladybeetles. *Advances in Genetics* **8**: 155–189.
- KOVACH, J. 2004. Impact of the multicolored Asian lady beetle as a pest of fruit and people. *American Entomologist* **50**: 165–167.
- KUZNETSOV, V.N. 1997. *Lady Beetles of Russian Far East*. Memoir No. 1. Center for Systematic Entomology and The Sandhill Crane Press, Inc., Gainesville, Florida, U.S.A.
- LAMANA, M.L. & MILLER, J.C. 1996. Field observations on *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae) in Oregon. *Biological Control* **6**: 232–237.
- LAMANA, M.L. & MILLER, J.C. 1998. Temperature-dependent development in an Oregon population of *Harmonia axyridis* (Coleoptera: Coccinellidae). *Environmental Entomology* **27**: 1001–1005.
- LOU, H.H. 1987. Functional response of *Harmonia axyridis* to the density of *Rhopalosiphum prunifoliae*. *Natural Enemies of Insects* **9**: 84–87.
- LUCAS, E., CODERRE, D. & VINCENT, C. 1997. Voracity and feeding preferences of two aphidophagous coccinellids on *Aphis citricola* and *Tetranychus urticae*. *Entomologia Experimentalis et Applicata* **85**: 151–159.
- MACK, R.N., SIMBERLOFF, D., LONSDALE, W.M., EVANS, H., CLOUT, M. & BAZZAZ, F.A. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. *Ecological Applications* **10**: 689–710.
- MAGRO, A., TENE, J.N., BASTIN, N., DIXON, A.F.G. & HEMPTINNE, J.L. 2007. Assessment of patch quality by ladybirds: relative response to conspecific and heterospecific larval tracks a consequence of habitat similarity? *Chemocoecology* **17**: 37–45.
- MAJERUS, M.E.N. 1994. *Ladybirds*. No. 81, New Naturalist Series. HarperCollins, London, U.K.
- MAJERUS, M., STRAWSON, V. & ROY, H. 2006. The potential impacts of the arrival of the harlequin ladybird, *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), in Britain. *Ecological Entomology* **31**: 207–215.
- MICHAUD, J.P. 2002. Invasion of the Florida citrus ecosystem by *Harmonia axyridis* (Coleoptera: Coccinellidae) and asymmetric competition with a native species, *Cycloneda sanguinea*. *Environmental Entomology* **31**: 827–835.
- MICHAUD, J.P. 2003. A comparative study of larval cannibalism in three species of ladybirds (Coleoptera: Coccinellidae). *Journal of Entomological Science* **28**: 92–101.
- NALEPA, C.A., KIDD, K.A. & AHLSTROM, K.R. 1996. Biology of *Harmonia axyridis* (Coleoptera: Coccinellidae) in winter aggregations. *Arthropod Biology* **89**: 681–685.
- OSAWA, N. 2000. Population field studies on the aphidophagous ladybird beetle *Harmonia axyridis* (Coleoptera: Coccinellidae): resource tracking and population characteristics. *Population Ecology* **42**: 115–127.
- OSAWA, N. 2011. Ecology of *Harmonia axyridis* in natural habitats within its native range. *BioControl* **56**: 613–621.
- PICKERING, G.J., KER, K. & SOLEAS, G.J. 2007. Determination of the critical stages of processing and tolerance limits for *Harmonia axyridis* for 'ladybug taint' in wine. *Vitis* **46**: 85–90.
- PICKERING, G.J., LIN, J.Y., RIESEN, R., REYNOLDS, A., BRINDLE, I. & SOLEAS, G. 2004. Influence of *Harmonia axyridis* on the sensory properties of white and red wine. *American Journal of Enology and Viticulture* **55**: 153–159.
- PICKERING, G.J., LIN, J.Y., SOLEAS, G., REYNOLDS, A., RIESEN, R. & BRINDLE, I. 2005. The influence of *Harmonia axyridis* on wine composition and aging. *Journal of Food Science* **70**: 128–135.
- PIMENTEL, D., LACH, L., ZUNIGA, R. & MORRISON, D. 2000. Environmental and economic cost of non-indigenous species in the United States. *Bioscience* **50**: 53–65.
- POUTSMA, J.A., LOOMANS, J., AUKEMA, M.B. & HEIJERMAN, T. 2008. Predicting the potential geographical distribution of the harlequin ladybird, *Harmonia axyridis*, using the CLIMEX model. *BioControl* **53**: 103–125.
- PRETORIUS, R.J., LOUW, S.vd M. & VENTER, P. 2010. New record of Aphididae (Hemiptera) and Coccinellidae (Coleoptera) species associated with shade-house-cultivated lettuce on a South African Highveld farm. *African Entomology* **18**: 365–368.
- SAINI, E. 2004. Presencia de *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae) en la provincia de Buenos Aires. Aspectos biológicos y morfológicos. *RIA* **33**: 151–160.
- SAKURATANI, Y., MARSUMOTO, Y., OKA, M., KUBO, T., FUJI, A. & UOTANI, M. 2000. Life history of *Adalia bipunctata* (Coleoptera: Coccinellidae) in Japan. *European Journal of Entomology* **97**: 555–558.
- SIMBERLOFF, D. 2004. Community ecology: is it time to move on? *American Naturalist* **163**: 787–799.
- SIMBERLOFF, D. & STILING, P. 1996. How risky is biological control? *Ecology* **77**: 1965–1974.
- STALS, R. & PRINSLOO, G. 2007. Discovery of an alien invasive, predatory insect in South Africa: the multi-coloured Asian ladybird beetle, *Harmonia axyridis*

- (Pallas) (Coleoptera: Coccinellidae). *South African Journal of Science* **103**: 123–126.
- STATISTICA. 2012. *Statistica 10 for Windows*. Statistica Enterprise System Technology, Tulsa, Oklahoma, U.S.A.
- STUART, R.J., MICHAUD, J., OLSEN, L. & MCCOY, C. 2002. Lady beetles as potential predators of the root weevil *Diaprepes abbreviatus* (Coleoptera: Curculionidae) in Florida citrus. *Florida Entomologist* **85**: 409–416.
- TAKAHASHI, K. 1989. Intra- and interspecific predations of lady beetles in spring alfalfa fields. *Japanese Journal of Entomology* **57**: 199–203.
- TATEM, A.J. 2009. The worldwide airline network and the dispersal of exotic species: 2007–2010. *Ecography* **32**: 94–102.
- VITOUSEK, P.M., D'ANTONIO, C.M., LLOYD, L., LOOPE, L.L., REJMANEK, M. & WESTBROOKS, R. 1997. Introduced species: a significant component of human-caused global change. *New Zealand Journal of Ecology* **21**: 1–16.
- VITOUSEK, P.M., D'ANTONIO, C.M., LOOPE, L.L. & WESTBROOKS, R. 1996. Biological invasions as a global environment change. *American Scientist* **84**: 468–487.
- WATANABE, M. 2002. Cold tolerance and myo-inositol accumulation in overwintering adults of a lady beetle, *Harmonia axyridis* (Coleoptera: Coccinellidae). *European Journal of Entomology* **99**: 5–9.
- WILLIAMSON, M. 1996. *Biological Invasions*. Chapman & Hall, London, U.K.
- YASUDA, H. & OHNUMA, N. 1999. Effect of cannibalism and predation on the larval performance of two ladybird beetles. *Entomologia Experimentalis et Applicata* **93**: 63–67.
- YASUDA, H., KIKUCHI, T., KINDLMANN, P. & SATO, S. 2001. Relationships between attacks and escape rates, cannibalism, and intraguild predation in larvae of two predatory ladybirds. *Journal of Insect Behavior* **14**: 373–384.
- YASUDA, H., TAKAGI, T. & KOGI, K. 2000. Effects of conspecific and heterospecific larval tracks on the oviposition behaviour of the predatory ladybird, *Harmonia axyridis* (Coleoptera: Coccinellidae). *European Journal of Entomology* **97**: 551–553.

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